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UNIVERSITY EXPERIMENTS WITH SAND FLY AND PELLAGRA..*S. J. Hunter.*

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University Experiments with Sand Fly and Pellagra.

An account of endeavors to substantiate the Sambon theory of the transmission of pellagra by the sand fly, *Simulium*.

BY S. J. HUNTER.

THE etiology of pellagra has been a subject of serious inquiry for over two hundred years. Formerly an old-world problem, it is now found widely distributed in this country. The date of its first appearance in North America is not well established.

Three theories have been advanced as to the cause of this disease: (1) The zeistic theory, based on the work of Ballardini, in 1845, giving corn poison due to the excessive use of corn products as the cause, supplanting the old theory of faulty metabolism; (2) the cottonseed products poison theory of Mizell, in 1911; and (3) the sand-fly theory of Sambon, dating from 1910. The first two rest on malnutrition; the last on the action of a parasite.

Since this paper deals only with the parasitic theory of Sambon, the grounds on which Sambon bases his theory may properly be outlined here:

A. The endemic centers of pellagra in Italy have remained the same since the disease was first described.

B. The season of the recurrence of pellagra coincides with the season of the appearance of the full-fledged sand fly, even to the extent that if the spring is early or late the sand fly is early or late in appearing, and pellagra cases are correspondingly early or late in their appearance.

C. In centers of pellagra infection whole families are attacked at times simultaneously.

D. In nonpellagrous districts the disease never spreads to others with the advent of a pellagrin from a pellagrous district.

E. In the case of a family which has removed from a pellagrous to a nonpellagrous district, the children born in the former district are pellagrins, while the children born subsequent to removal to a nonpellagrous district do not develop the disease.

F. The disease is not hereditary, although infants a few months old may become infected, especially if taken to the fields in pellagrous districts, where their mothers work during the season when sand flies are in evidence.

G. Pellagra is not contagious but is transmitted to each individual by an infected sand fly.[1]

In Kansas the latter part of July, 1911, the first authentic cases, those of a mother and child, were diagnosed by Dr. E. E. Liggett, of Oswego, the attending physician. Dean Crumbine, of the medical school of the University of Kansas and secretary of the State Board of Health, expressed an earnest desire to have the presence of the sand fly ascertained for that locality. Accordingly the author began, on August 1, a survey of the region. The streams were high and muddy from recent rains, so that adults were first sought. An extended opportunity which I enjoyed for study of this insect some years ago in the Mississippi valley, between Keokuk and Fort Madison, Iowa, rendered familiar the workings of the adult females on horses, especially young colts, on which after warm rains the attacks were so numerous and severe as to denude the animals' ears and throats, exposing patches of raw flesh.

At the beginning of this investigation there was only one *Simulium* in the University collection recorded from Kansas. The investigation then from the beginning has been prosecuted along three lines: (1) The survey on the distribution of the sand flies in Kansas; (2) a detailed investigation of their life habits and conditions under which they exist in Kansas; (3) their biting and feeding habits and the possibilities of inoculation.

The first two lines have been treated in the following paper (No. 9) by my graduate student, Mr. W. T. Emery, who has been my assistant throughout these experiments. The third

1. Sand-fly Transmission of Pellagra, The Journal A. M. A., Nov. 26, 1910, p. 1898.

subject requires the consideration of the digestive tract, and this Assistant Professor Hungerford has presented in paper No. 10, of this number.

The Thompson-McFadden Pellagra Commission, with headquarters at Spartanburg, S. C., is conducting an exhaustive survey into all possible factors which might reveal the cause of this disease, and the Bureau of Entomology is coöperating with this commission along entomological lines. The department of entomology at the University is concerned only with the entomological side of the question, and has thus far dealt only with the sand fly. The responsibility for the entomological side of the question rests with the author, and the pathologic side, as manifested by the monkeys subjected to the bite of the sand fly, rests with Dean Crumbine of the medical school. In this connection it might properly be noted that recently Doctor Harris has published an article in which he states he has been successful in producing pellagra[2] experimentally in monkeys. Based upon his experiments, then, the monkey becomes a susceptible animal.

The line of investigation followed was: transfusions on guinea pigs and monkeys, and transference of flies, exposed to the pellagrins, to monkeys and guinea pigs. In the experiments with each exposed animal there was a check or unexposed animal.

The transfusions and inoculations gave no positive results. Temperatures of the guinea pigs and monkeys were taken twice a day without any appreciable change. As this subject has been covered in a paper by Anderson and Goldberger,[3] who obtained similar results, we will pass to what we consider the more important phase of the Sambon theory, viz., the role of the sand fly.

His theory is protozoal, and, from analogy with the etiology of malaria, the parasite of pellagra in all probability would have to pass one stage in the body of its intermediate sand-fly host before it could resume its life in the human body. This hypothesis being true, transfusions obviously would be without results.

Taking up the sand fly, then, the only species found generally distributed in Kansas thus far is *Simulium vittatum,* as determined by Johannsen. *S. reptans* is the species referred to

2. Jour. A. M. A. Lx. No. 25, June 21, 1913, 1948-1949.

3. Public Health Rep., 1911, XXVL, 1003-1007.

by Sambon, and reported on this continent from Greenland only. Since several species of the genus *Anopheles* transmit malaria, it would seem permissible to consider another species belonging to the genus *Simulium* than that of *S. reptans,* referred to by Sambon. *S. vittatum* is distributed in Kansas, as shown by Emery in a map in the following paper.

For the location of the pellagrins we have Allen, Chautauqua, Montgomery, Labette, Shawnee, Sumner and Meade counties. All cases are in the midst of the sand-fly territory, except the Meade county case, and this man, a resident of that region for twenty years, spent the year of 1910 in the South. The Allen and Labette county cases are endemic. None of these patients have ever been out of the state, so it would seem that the cause now exists in the state.

For our experimental work in 1911, the Oswego pellagrin, a woman aged about 35, now in the second season of the disease, was used, and she very willingly did her part.

In all, 1282 live sand flies were used, and this phase of the work extended from August 21 to November 4. The plan was to divide the number of flies intended for each experiment into two lots, the one lot to be exposed to the pellagrin and then to the subject of experimentation, and the other exposed to the check.

Ten guinea pigs and two monkeys were used, and the temperature of all was taken morning and evening daily. The number of live flies exposed to the pellagrin and then to the guinea pigs was 499; the number of live flies exposed to the pellagrin and then the monkeys was 197. A part of those exposed to the pellagrin were reserved for fixation and sectional microscopic examination in the laboratory. Since only the females bite, the relative number of the sexes is important. In a count of 488 specimens, 219, or 42 per cent, were females.

Earlier in the season the flies did not seem to bite the patient, but beginning with October 12 they attacked her, biting freely, drawing the blood perceptibly from the pellagrin's arms. These flies were then divided, part placed in the flyproof cage with the male monkey; part with the guinea pigs. Repetitions of the same experiment were made almost daily during the stated period.

This closed on November 2, 1911, the work at Oswego, Kan., and it was transferred from Oswego to the University labora-

tory. On November 7 the male monkey used to receive inoculation became suddenly ill, growing flaccid and motionless, save for a high rate of respiration. He was later placed in charge of Doctor Boughten, pathologist, and Doctor Skoog, neurologist, of the University. He was chloroformed, autopsied, and the brain and spinal cord were studied by Doctor Skoog. This monkey was taken from the University hospital group. It now appears that some eighteen months before this monkey was exposed to poliomyelitis. While the time is remote, nevertheless the exposure obviously introduces a complication which will make it unsafe to depend on this instance, unless substantiated by many pure culture corroborations.

This closed the work for the year 1911, and as far as our knowledge goes is the first recorded instance of an endeavor to apply the Sambon theory in this manner.

The following year, 1912, the same method of work was conducted, and to avoid any possibility of contamination six monkeys were imported direct from India for this work.

The season previous the flies were very abundant. The season of 1912 they were notably rare, due principally to the flooded conditions of the creeks in the northern part of the state and improper amount of moisture in the southern part of the state. The almost total absence, therefore, of flies in July, as well as a want of pellagrins in the proper stage, prevented any successful inoculations at that time.

From the fall brood, however, inoculations were made without waiting for the flies to bite the monkeys, but securing inoculations through maceration attended by the proper aseptic precautions. The last inoculation was made on a female monkey on December 16, 17, 18, 1912, using flies that had emerged on the 12th and bitten the pellagrin on the 14th.

Work for 1913 was conducted under a special fund furnished by the Board of Educational Administration.

Special attention has been given this season to the biting habits of *Simulium* in nature. Heretofore we have experienced little difficulty in encouraging the sand fly to bite the patient, but no extended study had been given to the biting habits in nature.

During 1912 it was found that they would bite during the mornings and evenings, sometimes entering the tent. The season of 1913, however, was not favorable in Kansas for the development of the sand fly during the summer.

In August, therefore, this part of the study was transferred to Madison river, in the southern part of Montana, where all stages of the fly were unusually abundant. Here it was observed that the fly would bite the exposed parts, and was more active on cool days while the temperature was below 70° F.

Of special importance was the observation made by four members of the party that the bite of the fly was not always noticeable. For example, the writer sat through an entire evening meal in the tent with the sand fly biting on the face near the base of the nose. He was not aware of its presence there until informed at the close of the meal by his companions regarding the length of the time it had been there. The spot reddened in this case was about the size of a flax seed.

It seems probable also that it succeeds in attaching itself to the host through its mouth parts, because when once settled down to feeding it sticks to the host and is not readily detached.

Biting is not uniformly painless, as sometimes the insect could be detected by its first contact.

Regarding the morphology of the mouth parts, Mr. W. T. Emery, who has been my graduate student assistant in this work, has the paper following, dealing with that phase of the subject.

A second point to be here recorded is that the monkey, which we used all last year to receive inoculations from the sand fly, and which received its last inoculation on December 22, 1912, as recorded in my previous paper, late in November, 1913, began to show a marked stomatitis, accompanied by a diarrhœa. She has continued to lose in weight and the color of the face is changing from the normal to a pale ashy gray.

Summing up the evidence, then, in the work thus far for and against the Sambon theory:

1. The number of sand flies has been directly proportional to the number of cases of pellagra.

2. The first appearance of the cases of pellagra is coincident with the principal broods.

3. Just succeeding the time of the principal broods the flies appear to bite more vigorously.

4. Sand flies which have fed on human blood live several days longer than those which have not been so nourished, thus favoring an incubation period for a parasite, if such there be.

5. Pellagra, thus far in Kansas, has appeared almost entirely in one restricted locality. Of the nine cases recorded last year five were traced back to one town. In this region flies are widely distributed and unusually abundant.

6. No direct evidence has thus far been found which would in any way warrant any conclusion with reference to an association of the sand fly in the determination of the etiology of pellagra.

As far as our history of this subject goes, the appearance of pellagra in Kansas is of recent occurrence. Nearly all of the cases are those of natives that have never been out of the state. From this it would seem that the etiology of pellagra exists in Kansas.

The situation here is not complicated through long-standing conditions. It is the purpose, therefore, to continue this phase of the work, and with it the correlation of the clinical and pathological phases by the medical school of the University.

BIBLIOGRAPHY—THE SAND FLY AND PELLAGRA.

1910—Pellagra and Its Cause. Nature, vol. LXXXIII, p. 378.

LAVINDER, C. H. The Theory of the Parasitic Origin of Pellagra. Public Health Reports, vol. XXV, pp. 735, 736.

Sand-fly Transmission of Pellagra. Jour. Am. Med. Assn., vol. LV, p. 1898.

——— Recent Investigations on Pellagra. Nature, vol. LXXXIV, pp. 538, 539.

SAMBON, LOUIS W. Progress Report on the Investigation of Pellagra. Jour. Tropical Medicine and Hygiene, vol. XIII, pp. 271-282, 287-300, 305-315, 319-321.

KING, HOWARD D. The Etiologic Controversy Regarding Pellagra. Jour. Am. Med. Assn., vol. LIV, No. 11, p. 859.

SAMBON, L. W. Cause and Transmission of Pellagra. Policlinico, June, 1910. (Review, in Jour. Amer. Med. Assn., July, 1910.)

1911—HOWARD, L. O. *Simulium* and Pellagra. Report of entomologist for 1911, p. 34, U. S. Dept. Agri.

HEWITT, C. GORDON. *Simulium* Flies and Pellagra. Nature, vol. LXXXV, pp. 169, 170.

SHELFORD, R. *Simulium* and Pellagra. Nature, vol. LXXXV, p. 41.

CRUMBINE, S. J. Pellagra in Kansas. Bulletin State Board of Health, October, 1911, p. 190.

CARLETTI, M. V. Sand-fly Transmission of Pellagra. Gazetta degli Ospedali e delle Cliniche, Milan, May 28, 1911, XXXII, No. 64.

1911—JOHANNSEN, C. A. *Simulium* and Pellagra. Bul. 187, Maine Agricultural Exp. Station, January, 1911.

SAMBON, L. W. Progress Report of the Investigation of Pellagra, U. S. Dept. Agri. Ex. Station Record, vol. XXVI, abstract No. 8.

WALL, F. Recent Pellagra Investigations by the British Pellagra Commission. Jour. Trop. Med. and Hyg., December 15, 1911.

ROBERTS, S. R. Sambon's New Theory of Pellagra, and its application to conditions in Georgia. Jour. Amer. Med. Assn., vol. LVI, pp. 1713-1715.

THORINGTON, CHILTON. Some Suggestions as to the Etiology of Pellagra. Virginia Medical Semimonthly, vol. 16, p. 185.

BEALL, K. H. The Etiology of Pellagra. Jour. Am. Med. Assn., vol. LVII, p. 1683.

ALCOCK, A. Entomology for Medical Officers, pp. 123-131.

GRIMM, R. M. Pellagra: A Report on an Epidemiologic Study. Pub. Health Reports, vol. XXVII, No. 8, pp. 255-264.

CACCINI, A. Pellagra as we See it in Italy: Old and New Theories. Report of cases seen in New York City. Med. Record, Mar., 1911.

1912—HUNTER, S. J. The Sand Fly and Pellagra. Jour. Am. Med. Assn., vol. LVIII, pp. 547-548.

HUNTER, S. J. The Sand Fly and Pellagra. Jour. Econo. Ent., vol. V, pp. 61-63.

NICHOLLS, LUCIUS. Pellagra, "Sand Fly and Protozoön" versus "Maize" Theory. Jour. Trop. Med. and Hyg., vol. 15, No. 20, Oct. 15, 1912, pp. 305-306.

FORBES, S. A. On Black Flies and Buffalo Gnats (*Simulium*) as Possible Carriers of Pellagra in Illinois. 27th Rept. State Entomologist of Illinois, p. 21-52.

GARMAN, H. A Preliminary Study of Kentucky Localities in which Pellagra is Prevalent. Bull. 159, Ky. Agric. Exper. Station.

ROBERTS, STEWART R. Pellagra: History, Distribution, Diagnosis, Prognosis, Treatment, Etiology. C. V. Mosby Co., St. Louis.

NILES, GEORGE M. Pellagra. W. B. Saunders Co., Philadelphia.

RAVITCH, M. L. A Plea for an Earlier Diagnosis of Pellagra. Jour. Am. Med. Assn., vol. LIX, pp. 33-35.

WOOD, E. J. A Treatise on Pellagra. Appleton & Co., New York.

1913—HUNTER, S. J. Pellagra and the Sand Fly. Jour. Econom. Ent., vol. VI, No. 1, pp. 99-101.

ROBERTS, S. R. The Analogies of Pellagra and the Mosquito. Am. Jour. Med. Science, vol. CXLVI, No. 2, Aug., 1913, No. 479, p. 233.

JENNINGS, A. H., and KING, W. B. An Intensive Study of Insects as a Possible Etiologic Factor in Pellagra. From Am. Jour. of Med. Sciences, vol. 146, No. 3, p. 411.